Integration of Emerging Technologies Into Plant Germplasm Improvement Programs

Agricultural Research Service Workshop
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- C O N T E N T S -

I - WORKSHOP PARTICIPANTS

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G. W. Burton USDA ARS SR Forage & Turf Research Georgia Coastal Plain Expr Sta Tifton, GA 31793 Telephone: 912/386-3353	Genetic improvement of grass and forage crops. Improved methods of handling germplasm and breeding cross-pollinated species.
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J. P. Helgeson USDA ARS NCR Disease Resistance Laboratory Dept of Plant Pathology University of Wisconsin Madison, WI 53708 Telephone: 608/262-1541	Genetic modification of crop plants. Molecular mechanisms of disease resistance. Plant tissue and cell culture.
R. R. Hill USDA ARS NER U.S. Regional Pasture Research Laboratory Curtin Road University Park, PA 16802 Telephone: 814/237-7683	Improved germplasm of alfalfa. Genetics of autotetraploids. Quantitative genetics. Improved breeding methodology.
A. O. Jackson Dept of Botany & Plant Pathology Purdue University West Lafayette, IN 47907 Telephone: 317-494-4600	Structure, function, and replication of the genome of plant viruses. Development of disease caused by plant viruses.

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R. J. Kohel USDA ARS SR Cotton Genetics Research P. O. Drawer DN College Station, TX 77840 Telephone: 713/846-8821, x-311	Improved germplasm of cotton. Management of plant germplasm resources. Qualitative genetics. Selection criteria for physiological traits.
T. J. McCoy USDA ARS WR Alfalfa Production Research College of Agriculture University of Nevada Reno, NV 89557 Telephone: 702/784-5336	Improved germplasm of alfalfa. Interspecific gene transfer. Cell and tissue culture. Cytogenetics.
N. C. Nielsen USDA ARS NCR Plant Science Research Lilly Hall, Life Sciences Bldg. Purdue University W. Lafayette, IN 47907 Telephone: 317/494-8057	Improved soybean germplasm. Plant molecular biology. Genetic modification of seed protein.
D. R. Pring USDA ARS SR Plant Science Research Univ of Florida, Bldg 164 Agronomy & Physiology Bldg. Gainesville, FL 32611 Telephone: 904/392-6180	Cytoplasmic inheritance of male sterility and disease susceptibility in maize, sorghum, and millet. Molecular biology. Transcription sequences affecting traits.

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J. N. Rutger USDA ARS WR Dept of Agronomy and Range Science University of California Davis, CA 95616 Telephone: 916/752-2460	Improved germplasm of rice. Induced mutation. Interspecific hybridization. Male sterility.
J. M. Stewart USDA ARS SR Cotton Physiology Research 310 Plant & Soil Science Bldg University of Tennessee Knoxville, TN 37916 Telephone: 615/974-8862	Developmental physiology and morphology of cotton. Interspecific hybridization. Embryo rescue and culture.
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II - EXECUTIVE SUMMARY

Projected long-term domestic and foreign demands for the production of our major field, horticultural, range, and forage crops cannot be met with current yields and productivity. The foundation of crop productivity is the inherent genetic potential of the plants themselves. Although gains in yields of major crop plants in the United States have been estimated as currently averaging about one percent per year, this rate of gain falls short of meeting the projected needs of the 21st Century. We must increase productivity (yield per unit of resource input) if we are to provide consumers with food and fiber at reasonable prices, conserve natural resources for sustained production, and compete in world markets. This goal can be accomplished most efficiently by increasing the rate of improvement of plant germplasm. The objective of this workshop was to identify the science and the priority research approaches needed to develop the information and technology which will accelerate the improvement of germplasm of the higher plants.

The information generated will be of value to scientists and administrators of the entire agricultural community. It is recognized that the priorities among specific research approaches needed will vary among commodities, species, and production regions. Therefore this report is being distributed to appropriate commodity group scientists for use as a framework to assess specific research needs for the respective commodities. The responses from these groups will help guide ARS' long-range and operational planning.

Four major research goals were identified. These goals and the research approaches needed to achieve each goal are:

Goal A - Develop basic knowledge concerning the organization of plant genomes, gene structure, and the regulation of gene expression.

Research Approaches to Achieve Goal A

A-1 Determine how genetic information is organized in plant genomes.
A-2 Elucidate the organization and structure of plant genes.
A-3 Determine how genes are regulated.
A-4 Develop methods to identify and isolate genes of interest.

Goal B - Develop more efficient methods for the identification and transfer of desirable genes within and between species.

Research Approaches to Achieve Goal B

B-1 Develop more efficient methods for the production and utilization of unique hybrids.
B-2 Increase knowledge about the mechanisms and efficiency of genetic recombination.
B-3 Develop procedures to more efficiently produce and utilize mutations.
B-4 Increase knowledge, efficiency, and effectiveness of methods for cell culture and plant regeneration.
B-5 Develop vector-mediated gene transfer technology.

Goal C - Develop more effective and efficient selection methods for modifying traits associated with genetic improvement of plants.

Research Approaches to Achieve Goal C

C-1 Increase efficiency of selection procedures for improvement of important agronomic traits.
C-2 Develop selection criteria for enhancing productivity in stress environments.
C-3 Develop screening procedures to detect genetic changes in physiological and biochemical processes that contribute to productivity.
C-4 Develop methods to screen for desired traits in modified organs and tissues.
C-5 Devise approaches for selection of useful traits in callus culture or at the cellular level.

Goal D - Develop more effective reproductive and propagation systems.

Research Approaches to Achieve Goal D

D-1 Develop better understanding of basic reproductive biology of plants.
D-2 Manipulate modes of reproduction and propagation to develop and evaluate new gene combinations.
D-3 Utilize novel reproductive methods to propagate new cultivars.
D-4 Develop more efficient methods of cytoplasmic and chromosomal manipulations to develop new gene combinations.
D-5 Develop cell culture as a reproductive system for producing or maintaining new genetic combinations.

These identified goals and research approaches emphasize the importance of integrating the emerging technologies of genetic engineering into ongoing plant germplasm improvement programs. New tools and more efficient methods of genetic modification are essential if we are going to meet future crop production needs. Plant breeding will continue to be the delivery system for these new technologies.

Detailed descriptions of each Research Approach with comments on priorities among the approaches and on the resources needed to implement the research are presented in the following report.

III - WORKSHOP REPORT

Problem Statement

Projected long-term domestic and foreign demands for the production of our major field, horticultural, range, and forage crops cannot be met with current yields and productivity. The foundation of crop productivity is the inherent genetic potential of the plants themselves. Although gains in yields of major crop plants in the United States have been estimated as currently averaging about one percent per year, this rate of gain falls short of meeting the projected needs of the 21st Century. We must increase productivity (yield per unit of resource input) if we are to provide consumers with food and fiber at reasonable prices, conserve natural resources for sustained production, and compete in world markets. This goal can be accomplished most efficiently by increasing the rate of improvement of plant germplasm.

We do not understand the genetic structure and function of higher plants well enough to fully exploit existing technologies for plant germplasm improvement. We have made only limited progress in integrating emerging genetic engineering technologies into existing programs. Therefore, we need to strengthen our efforts to create a research environment that adequately fosters the discovery of new and potentially useful tools and methods for germplasm improvement. The development of such a program will enable us to more efficiently and effectively modify quantitative and qualitative traits that control yield potential; product quality; resistance to pests; tolerance to environmental stresses; and adaptation to economical cultural, harvesting, handling, and management practices.

The objective of this workshop was to identify the science and the priority research approaches needed to develop the information and technology which will accelerate the improvement of germplasm of higher plants.

Scope of Workshop

Research in new areas of genetic engineering is dependent on genes in existing germplasm collections. The collection, maintenance, evaluation, and information management of plant germplasm resources, however, are being considered in other workshops. Thus, this workshop concentrated on the development of methods and technologies to 1) identify, produce, and isolate useful genetic variation and 2) utilize that variation to improve germplasm. Considerations were restricted to the higher plants, but included the entire range of current and potential technologies applicable to both cellular and whole-plant populations. It is recognized that the priorities among specific research approaches needed will vary among commodities, species, and production regions. Therefore, the objective of this workshop was limited to identifying the overall science and broad approaches needed for germplasm improvement. As the next step, this report is being distributed to appropriate commodity group scientists to use as a framework to assess specific research needs for the respective commodities. The responses from these groups will help guide ARS' long-range and operational planning.

Procedures

Step 1 - The entire group of workshop participants devoted the first half-day session to the identification of barriers to the development of more efficient germplasm improvement procedures. These barriers were organized into four subgroups and stated in the form of research goals. Workshop participants were then divided into four teams of six scientists each to follow through with Steps 2, 3, and 4.

Step 2 - Working in team groups and plenary session, participants identified the research approaches needed to achieve each research goal.

Step 3 - Each research approach was described in further detail to enable administrators to determine the specific resources and management structures needed to implement the research.

Step 4 - Working in team groups and plenary sessions, participants recommended priorities and balance among the research approaches identified to achieve each goal.

Research Needed to Accelerate Improvement of Germplasm

The participants identified four major research goals required to accelerate improvement of germplasm of higher plants. These goals, the research approaches needed to achieve each goal, and the recommended priorities and balance among approaches are outlined below:

GOAL A

Statement of Goal

Develop basic knowledge concerning the organization of plant genomes, gene structure, and the regulation of gene expression.

Research Approaches to Achieve Goal

A-1 Determine how genetic information is organized in plant genomes.
A-2 Elucidate the organization and structure of plant genes.
A-3 Determine how genes are regulated.
A-4 Develop methods to identify and isolate genes of interest.

Priorities and Balance Among Approaches

To modify germplasm we need increased knowledge of the plant genome, including genetic, cytogenetic, breeding, and molecular biology approaches. The acquisition of these data will allow a better understanding of the genome, how genes are assembled, and how they function and are regulated. Molecular biology offers intriguing possibilities in acquiring basic knowledge in these areas, and integration with genetic areas may ultimately lead to the capability of selectively modifying and expressing genes of interest.

We place highest priority on the integration of molecular biology with physiology, genetics, and breeding to study the regulation and control of specific genes (A-3). This will require judicious selection of traits and species and extensive coordination among a number of disciplines. The large resource requirement and the uncertainty of results dictate the selection of specific model systems rather than a broad approach. We further recognize the prerequisite to understand organization and information in plant genomes (A-1) and organization and structure of genes (A-2), and to devise more efficient means for identifying and isolating genes (A-4). This research will provide a background of fundamental information useful in the design of strategies to selectively modify plants.

GOAL B

Statement of Goal

Develop more efficient methods for the identification and transfer of desirable genes between and within species.

Research Approaches to Achieve Goal

B-1 Develop more efficient methods for the production and utilization of unique hybrids.
B-2 Increase knowledge of the mechanisms and efficiency of genetic recombination.
B-3 Develop procedures to more efficiently produce and utilize mutations.
B-4 Increase knowledge, efficiency, and effectiveness of methods for cell culture and plant regeneration.
B-5 Develop vector-mediated gene transfer technoogy.

Priorities and Balance Among Approaches

Elucidation of the mechanisms involved with recombination (B-2) appear to be of paramount importance since these are the primary constraints that limit modification of current breeding practices. An understanding of these phenomena should facilitate improvement of procedures already well developed.

Research directed to the acquisition of regeneration protocols for the economically important crop species (B-4) and the development of vectors useful for the *in vitro* manipulation of DNA and its insertion into cells (B-5) are of equal importance. However, the low probability of their making a profound immediate effect on crop improvement techniques prompts us to ascribe slightly less priority to these approaches than those of B-2. It should also be pointed out that, while approaches B-4 and B-5 have considerable direct potential impact on crop improvement, success in this area will greatly facilitate experimental studies on the regulation of gene expression (Goal A), and vice-versa. For maximum progress, it is important to closely coordinate the research in approaches B-4 and B-5 with the research as set out in Goal A.

Approach B-1 would lead to improvement of current methods for crop improvement, although we feel that the technology in this area is reasonably advanced. Mutation breeding (B-3) has been of limited use in previous efforts toward crop improvement. While induced mutations could serve an increased role in providing new genetic variability, the most significant gains would appear to be possible after effective means to selectively modify given traits have been developed, and these may be facilitated by development of approaches B-4 and B-5.

GOAL C

Statement of Goal

Develop more effective and efficient selection methods for modifying traits associated with genetic improvement of plants.

Research Approaches to Achieve Goal

C-1 Increase efficiency of selection procedures for improvement of important agronomic traits.

C-2 Develop selection criteria for enhancing productivity in stress environments.

C-3 Develop screening procedures to detect genetic changes in physiological and biochemical processes that contribute to productivity;

C-4 Develop methods to screen for desired traits in modified organs and tissues.

C-5 Devise approaches for selection of useful traits in callus culture or at the cellular level.

Priorities and Balance Among Approaches

The current average rate of increase in yield across most crop species is one percent per annum; several crop species have reached a yield plateau. This rate is a measure of the effectiveness and efficiency of selection systems currently utilized in plant improvement.

The challenge offered to Goal C is to increase that one percent per annum rate. It is essential to identify useful genes and develop specific and reliable ways of incorporating them into plants, preferably without disturbing the other characteristics of the cultivar. Here is where the new and the current techniques indeed interface. This requires both improvement of current selection criteria and development of effective ways to employ new technologies. Increased research is needed both in basic and applied areas. While we recognize the need to adequately support all of these approaches, the consensus of priorities is: C-1 (highest), C-2 and C-5 (tied for second), C-3 (third), and C-4 (fourth).

GOAL D

Statement of Goal

Develop more effective reproductive and propagation systems.

Research Approaches to Achieve Goal

D-1 Develop better understanding of basic reproductive and propagation systems.
D-2 Manipulate modes of reproduction and propagation to develop and evaluate new gene combinations.
D-3 Utilize novel reproductive methods to propagate new cultivars.
D-4 Develop more efficient methods of cytoplasmic and chromosomal manipulations to develop new gene combinations.
D-5 Develop cell culture as a reproductive system for producing or maintaining new genetic combinations.

Priorities and Balance Among Approaches

Research on control of modes of reproduction and propagation can significantly increase crop productivity. For example, production and utilization of sorghum increased dramatically after research on sterility control changed breeding from self- to cross-pollinated crop procedures. The control of apomixis in forage grasses and other new genetic methods offer almost unlimited potential for changing reproductive systems and the economics of cultivar development and use.

A balance between basic and applied research over a range of crop species is needed. Determination of priorities, balance, and timing among research approaches vary with species and require input from ARS and State research scientists, industry representatives, and crop advisory committees on a crop-by-crop basis.

Multidisciplinary interaction via formation of research teams (potentially including state, federal, and industry personnel) at one location and via strong linkages and communication among research scientists at different locations is strongly encouraged.

Further Description of Research Approaches

See Appendix for a further description of each Research Approach with comments on the resources and organizational structures needed to achieve goals.

IV - APPENDIX
FURTHER DESCRIPTION OF RESEARCH APPROACHES

Goal A - Research Approach A-1

Research Approach
Determine how genetic information is organized in plant genomes.

Further description of approach
To enable a better understanding of plant genome information and architecture through genetic, cytogenetic, and molecular means. Examples of research include: a) chromosome morphology and aberrations; b) gene mapping, conventional and contemporary; and c) determination of molecular bases of genomic instabilities.

Short-term or long-term research
Long-term with short-term benefit.

Disciplines or scientific expertise needed
Genetics, cytogenetics, molecular biology.

Special facilities and equipment needed
Microscopy, centrifugation, flow cytometry, cloning laboratories.

Environmental requirements
Genetic and cytogenetic studies for certain species must be conducted in areas where the plant is adapted.

Immediate "research users"
Basic plant scientists, industry, molecular biologists, breeders, and geneticists.

Organizational structure needed
Coordination, and linkage among disciplines.

Goal A - Research Approach A-2

Research Approach
Elucidate the organization and structure of plant genes.

Further description of approach
Basic studies on genes and flanking regions; primary sequence; control mechanisms; introns; promotors; base modification (methylation).

Short-term or long-term research
Long-term, short-term advances; others long-term, continual.

Disciplines or scientific expertise needed
Molecular biology.

Special facilities and equipment needed
Standard molecular biology laboratories.

Environmental requirements
None.

Immediate "research users"
Basic plant scientists and molecular biologists.

Organizational structure needed
Critical mass; ability to interact with similar projects, shared instrumentation in some cases. Coordination.

Goal A - Research Approach A-3

Research Approach
Determine how genes are regulated.

Further description of approach
A major problem facing modification of germplasm is understanding the nature of regulation of gene expression. Examples include research in: a) developmental gene regulation, b) multigenic traits and gene interaction, c) responses to environmental and biological (insects, diseases, microorganisms) stress, d) heterosis, e) nuclear-cytoplasmic interactions, f) biochemical pathways and their regulation, g) genotype-environment interactions, and h) temporal gene regulation.

Short-term or long-term research
Some short-term; others long-term.

Disciplines or scientific expertise needed
Physiology, biochemistry, pathology, genetics, molecular biology.

Special facilities and equipment needed
Standard molecular biology laboratories, environmental control facilities, and instrumentation.

Environmental requirements
Some commodity requirements.

Immediate "research users"
Physiologists and breeders.

Organizational structure needed
Provide for multidisciplinary interactions and communication.

Goal A - Research Approach A-4

Research Approach
Develop methods to identify and isolate genes of interest.

Further description of approach
A limited number of plant genes have been isolated and characterized. New strategies must be designed for complex, multigenic traits and to identify and isolate genes for which no product is known. Genetic, physiological, biochemical, and molecular approaches are needed to acquire knowledge of gene structure and regulation, which would lead to more efficient manipulation of germplasm. Existing methods of gene product isolation and characterization need to be extended to complex gene functions. Examples of research areas are: a) physiology and biochemistry of complex traits; b) gene isolation via controlling element insertional inactivation and new gene isolation strategies; c) source materials including mutants and isogenic lines; and d) monogenic or simply inherited traits such as disease resistance, herbicide sensitivity, insecticide sensitivity, etc.

Short-term or long-term research
Significant short-term advances; other long-term.

Disciplines or scientific expertise needed
Genetics, physiology, biochemistry, molecular biology, pathology, and entomology.

Special facilities and equipment needed
Broad.

Environmental requirements
Some commodity requirements.

Immediate "research users"
Breeders, industry, basic plant scientists, wide range of basic-applied scientists.

Organizational structures needed to facilitate research
Multidiscipline interactions, coordination, and communication.

Goal B - Research Approach B-1

Research Approach
Develop more efficient methods for the production and utilization of unique hybrids.

Further description of approach
Research for the required methodology includes chemical and environmental manipulation to improve hybridization efficiency, chromosome doubling, haploidization, in vitro fertilization, embryo rescue, species bridging techniques, etc. Research should be conducted to provide a more complete description of the fundamental genetic, morphological and biochemical phenomena involved in the reproductive process. We emphasize the importance of continuing to improve current technologies for hybrid production and utilization.

Short-term or long-term research
Hybrid production will have both short- or long-term benefits to crop improvement depending upon the species involved. A basic understanding of reproductive biology will have increased impact over a long period of time as additional information becomes available.

Disciplines or scientific expertise needed
Botany, physiology, genetics, cytogenetics, and biochemistry.

Special facilities and equipment needed
None(germplasm resources).

Environmental requirements
There may be specific instances where environmental effects assume importance in the production and/or selection of specific hybrids.

Immediate "research users"
Applied and basic scientific community.

Organizational structure needed
A coordinated effort will be required by the full spectrum of biological disciplines, but individual researchers could work efficiently providing communication with other areas of expertise.

Goal B - Research Approach B-2

Research Approach
Increase knowledge of genetic recombination and develop methods to improve its efficiency.

Further description of approach
Provide understanding of the fundamental mechanisms of genome organization, synapsis and recombination. Supply a thorough description of the cytogenetics of available germplasm. For closely related genomes, develop more efficient ways of using current methods such as backcross, recurrent, and pedigree

selections. For distantly related genomes develop methodology to induce fertility, discourage genomic isolation, and promote recombination in hybrids. In both cases research should be directed toward reducing the time required to generate desired recombinants, increasing the accuracy of recognizing them, and increasing the efficiency of their recovery. Research should be conducted to enable recognition of genes or gene blocks going into and coming out of hybrid combinations. We emphasize the importance of continuing to improve current technologies for achieving genetic recombination.

Short-term or long-term research
Recombinants can be obtained which will be of immediate use. Knowledge of the basic mechanisms involved will guide development of increasingly efficient technologies.

Disciplines or scientific expertise needed
Cytology, cytogenetics, population and quantitative genetics, physiology, biochemistry, molecular biology.

Special facilities and equipment needed
Microscopic; micromanipulation equipment.

Environmental requirements
There may be instances where environmental effects may assume importance, especially in those cases where whole plants must be grown for observation and selection.

Immediate "research users"
Applied and basic scientific community.

Organizational structure needed
Team efforts will be required. The teams will include molecular biologists, physiologists, geneticists, plant breeders, and cytogeneticists. The teams should be in close communication with workers engaged in fundamental medical and animal research where results for any area can immediately impact plant work.

Goal B - Research Approach B-3

Research Approach
Develop procedures to more efficiently produce and utilize mutations.

Further description of approach
Provide improved chemical and radiological approaches to generate genetic variability useful for modification of plants that can not be improved by conventional breeding techniques, or to create useful traits without modifying desirable rare gene combinations. Conduct research to develop new techniques for site directed mutations either *in vitro* (e.g., recombinant techniques) or *in vivo* to produce new genetic combinations. These combinations could be at the micro (e.g., point mutation) or macro scale (e.g., chromosome rearrangements and positional effects). Research is required to improve the efficiency and accuracy for detecting, maintaining, and evaluating new genetic structures.

Short-term or long-term research
Short-term gains can be realized from mutants generated and selected in response to specific needs (disease resistance, quality, etc.), but long-term efforts will be required to develop efficient methods.

Disciplines or scientific expertise needed
Geneticists, plant breeders, and molecular biologists will be the principle practitioners. Consultation with biochemists and physiologists will be required.

Special facilities and equipment needed
Usually not needed for generation of mutants, but may be needed for selection of specific recombinants.

Environmental requirements
Special environments for selection of specific recombinants may be needed.

Immediate "research users"
Applied plant breeders and geneticists.

Organizational structure needed
Standard approaches to mutation breeding would require applied breeders and geneticists. Applications which involve directed mutation in vitro and in vivo will require coordinated effort by molecular biologists, physiologists, biochemists, and plant geneticists.

Goal B - Research Approach B-4

Research Approach
Increase knowledge, efficiency, and effectiveness of methods for cell culture and plant regeneration.

Further description of approach
Reliable protocols must be developed for the culture of cells from diverse species and types of tissue. Improved methods of general application are also needed for the preparation of protoplasts, cell fusion, and transfer of organelles and chromosomes. The genetic and biochemical principles involved in regeneration of plants must be determined. In addition the recognition and selection of recombinants or transformed cells require improved methodology. We recommend that this work be carried out using crops of major economic importance.

Short-term or long-term research
Most of the researh is anticipated to be long-term, but progress along the way will have immediate applications in the utilization of somaclonal variation, somatic hybridization, and molecular genetics for crop improvement.

Disciplines or scientific expertise needed
Physiology, biochemistry, cytogenetics, genetics, molecular biology, and pathology.

Special facilities and equipment needed
Micromanipulators, electrofusion equipment, and tissue culture facilities.

Environmental requirements
No restrictions initially, although specific environments may be required for screening applications.

Immediate "research users"
Breeders, geneticists, and basic plant scientists.

Organizational structure needed
Direct association with plant improvement programs is essential. Close communication between researchers on different commodities is necessary. A team approach on the various aspects simultaneously would be desirable.

Goal B - Research Approach B-5

Research Approach B-5
Develop vector mediated gene transfer technology.

Further description of approach
Construct effective vectors (modified plasmids such as crown gall, chromosomes, nuclei, transposable elements, viruses, etc.) for the insertion of new genetic information into cells. Research is needed to assess the feasibility of transforming plants by direct injection of vectors into isolated cells or intact plants by fusion of loaded liposomes with plant protoplasts, by the use of electricity to reduce permeability of membranes, etc. Protocols should be developed which permit introductions of DNA at specific sites in the genome and permit evaluation of positional effects. Included in this technology will be development of probes to identify specific genes or gene products. While the initial objective of the research should be to delineate the basic principles involved in gene transfer and expression, efforts to adapt the methodologies to improvement of crops with major economic importance should be made at the earliest possible opportunity.

Short-term or long-term research
Short-term benefits to crop improvement will probably be limited to development of ways to rapidly and accurately identify desirable genetic variability. Long-term benefits will have an impact both as a new tool for practical plant breeding and as an opportunity to alter existing genetic information, to construct new combinations, and to introduce new DNA from diverse sources. The research may provide insight into basic genetic processes presently not understood such as durable resistance, genetic switching mechanisms, etc.

Disciplines or scientific expertise needed
Initial participants will be molecular biologists, biophysicists, biochemists, geneticists, and physiologists. Breeders will be required to adapat these techniques to practical breeding efforts and to identify goals.

Special facilities and equipment needed
None for the development of basic vehicles. Micromanipulation equipment, DNA sequencer, microprotein sequencer.

Environmental requirements
Special requirements may become necessary for specific recombinant selections.

Immediate "research users"
Mainly basic plant scientists. Methodologies such as recombinant DNA used as probes for specific genotypes could be used by practical breeders.

Organizational structure needed
Interdisciplinary team efforts will be required. Initial research will require geneticists, molecular biologists and biochemists. Application of techniques will require collaboration between breeders, geneticists, physiologists, and molecular biologists.

Goal C - Research Approach C-1

Research Approach
Increase efficiency of selection procedures for improvement of important agronomic traits.

Further description of approach
Improve efficiency of modifying those factors under genetic control that limit crop productivity and quality of product including disease and insect resistance and response to stress. Fundamental research in the genetics and population dynamics of pathogens and insects, the genetic control of host plant-pest interactions, and the genetic control of response to environmental stress is basic to developing new and innovative screening techniques to increase efficiency of plant improvement programs.

Short-term or long-term research
Short-term: Maintain the current level of genetic advance in economic crop plants; and long-term: Improve the rate of gain per unit of research resources.

Disciplines or scientific expertise needed
Plant pathology, genetics, physiology, and entomology.

Special facilities and equipment needed
None.

Environmental requirements
Restrained to areas of primary centers of adaptation of crop species.

Immediate "research users"
Plant breeders and geneticists, industry R&D, and commodity producers.

Organizational structure needed
Team efforts combining relevant disciplines are essential to progress.

Goal C - Research Approach C-2

Research Approach
Develop selection criteria for enhancing productivity in stress environments.

Further description of approach
None.

Short-term or long-term research
Short-term and long-term.

Disciplines or scientific expertise needed
Genetics, plant physiology, biochemistry, microclimatology, and soil science.

Special facilities and equipment needed
In general, standard equipment. Some special facilities and equipment may be required, depending upon the stress studied.

Environmental requirements
Locations where stresses occur and the availability of controlled environments.

Immediate "research users"
Applied public and private plant breeders.

Organizational structure needed
Team efforts with extensive linkages with other scientists.

Goal C - Research Approach C-3

Research Approach
Develop screening procedures to detect genetic changes in physiological and biochemical processes that contribute to productivity.

Further description of approach
1) Develop a better understanding of physiological and biochemical processes; 2) Relate processes and their interrelationships to plant growth (productivity); and 3) Determine inheritance patterns and develop screening or selection criteria.

Short-term or long-term research
Long-term: A sustained effort is needed. To date, research programs have declined after initial failures to achieve immediate success.

Disciplines or scientific expertise needed
Plant genetics, physiology, biochemistry, entomology, and pathology.

Special facilities and equipment needed
Standard laboratories. New instrumentation will be used as it becomes available.

Environmental requirements
Controlled environments (growth chambers) are essential. Field environment will depend on crop involved.

Immediate "research users"
Intermediate results will be used by other basic scientists. Applied plant breeders will apply screening procedures (final product) as they are developed.

Organizational structure needed
Team efforts (centers of excellence) are essential. Plant geneticists should be a part of team from beginning.

Goal C - Research Approach C-4

Research Approach
Develop methods to screen for desired traits in modified organs and tissues.

Further description of approach
Develop methods to assay for resistances to pests, toxins, and unfavorable environments. Develop molecular probes for altered genes and gene products. Develop screens for altered cells in presence of unaltered cells. Develop methods for separating altered and unaltered cells.

Short-term or long-term research
In the short term, this research could provide improved assays for desired traits in whole plants and for mutant cells. In the long term, this research will aid in selecting modified plants from callus cultures and other modified or variant tissues.

Disciplines or scientific expertise needed
Biochemists, molecular biologists, plant physiologists, virologists, entomologists, plant geneticists, and plant breeders will be required.

Special facilities and equipment needed
Specialized environmental control rooms, "state-of-art" spectrometers, and other equipment for molecular biologists will be required in addition to standard laboratory facilities.

Environmental requirements
None. Proximity to where crop is grown would be useful.

Immediate "research users"
Initially basic plant scientists will use these methods. When sufficiently developed and validated by correlations with intact plants, applied plant breeders could use the methods

Organizational structure needed
An interdisciplinary approach will be required.

Goal C - Research Approach C-5

Research Approach
Devise approaches for selection of useful traits in callus culture or at the cellular level.

Further description of approach
Methodology for efficient regeneration of protoplasts into intact plants needs to be developed for the major crop species. Selective culture media need to be developed to identify and isolate mutants with resistance or tolerance to toxins, herbicides, excess salinity, and heavy metals. Overproduction of desired gene products such as nutrients, antibiotics, or toxins detrimental to

pests may also be detected on selective culture media. Development of assay methods for identification of gene products is crucial for selection of modified cultures.

Short-term or long-term research
Research approach is long-term with at least a 5-year to a decade lag time anticipated before intact plants become available for genetic analysis or breeding programs.

Disciplines or scientific expertise needed
Individual with experience in tissue culture, microbial genetics, molecular biology, plant physiology, and plant morphology should form the core of a research team. Input from geneticists, plant pathologists, entomologists, nematologists, and microbiologists will expedite selection for various traits.

Special facilities and equipment needed
Special facilities are not needed, but adequate equipment is critical to achieving goals, and the essential equipment is expensive.

Environmental requirements
None.

Immediate "research users"
Plant geneticists who will determine heritibility of selected traits and whether these traits are suitable for crop improvement. Plant breeders subsequently will incorporate traits into suitable agronomic backgrounds.

Organizational structure needed
Teamwork is an essential factor in efficient exploitation of cell techniques. The routine work is especially labor intensive, requiring considerable effort from persons requiring little technical training.

Goal D - Research Approach D-1

Research Approach
Develop better understanding of basic reproductive biology of plants.

Further description of approach
This research will result in information on plant reproductive morphology, biochemical and physiological control of reproductive processes, and inheritance of morphological and physiological variants in the reproductive process.

Short-term or long-term research
Both short- and long-term researh is required. Information on reproductive morphology may be available or readily obtained. Identifying and understanding variability in reproductive processes will be a long-term goal.

Disciplines or scientific expertise needed
Morphology, genetics (all aspects including cytogenetic and molecular), physiology, and biochemistry.

Special facilities and equipment needed
None.

Environmental requirements
No restrictions.

Research users
Plant scientists and applied plant breeders.

Organizational structure needed
Independent research with interdisciplinary communications.

Goal D - Research Approach D-2

Research Approach
Manipulate modes of reproduction and propagation to develop and evaluate new gene combinations.

Further description of approach
Additional research is needed to ensure that no plant breeder is limited to the common modes of reproduction for the crop on which he works. Examples include: 1) control of sterility and fertility systems; 2) use of 2N gametes; 3) manipulation of apomixis; 4) use of anther culture; 5) transfer of cytoplasmic genomes; 6) meristem culture to eliminate seed-borne pathogens; and 7) vegetative propagation of rare genotypes when necessary.

Short-term or long-term research
Both short- and long-term researh are required.

Disciplines or scientific expertise needed
Biochemistry, physiology, genetics, cyto[illegible]tics, morphology, and pathology.

Special facilities and equipment needed
None.

Environmental requirements
No restrictions.

Research users
Unlimited

Organizational structure needed
Team effort--not necessarily at the same location.

Goal D - Research Approach D-3

Research Approach
Utilize novel reproductive methods to propagate new cultures.

Further description of approach
Same as D-2 except on an applied production scale. Areas of research include control of sterility and fertility systems through genetic, chemical, or physical means; use of meristem culture to eliminate seed-borne pathogens; manipulation of apomixis; vegetative propagation of rare genotypes; use of 2N gamete techniques; use of anther culture techniques; and transfer of cytoplasmic genomes.

Short-term or long-term research
Both short- and long-term researh is required.

Disciplines or scientific expertise needed
Genetics, cytogenetics, physiology, morphology, pathology, biochemistry, economics, engineering, and extension.

Special facilities and equipment needed
Some production scale facilities, as opposed to laboratory scale equipment, may be required.

Environmental requirements
Crop production region.

Immediate "research users"
Applied plant breeders and farmers.

Organizational structure needed
ARS, state, and industry cooperation is needed.

Goal D - Research Approach D-4

Research Approach
Develop more efficient methods of cytoplasmic and chromosomal manipulation for new gene combinations.

Further description of approach
Change (increase or decrease) in chromosome numbers and ploidy levels; create chromosome substitution or addition lines; change pairing relationships; modify chromosome structures in order to control inheritance of specified genes; and manipulate cyptoplasmic factors controlling fertility and sterility.

Short-term or long-term research
Both short- and long-term research included.

Disciplines or scientific expertise needed
Genetics (all aspects), cytogenetics, physiology, biochemistry, and morphology.

Special facilities and equipment needed
Electron microscope in some laboratories. Equipment for some techniques cannot be determined at present.

Environmental requirements
No restrictions.

Immediate "research users"
Other plant scientists.

Organizational structure needed
Depends on nature of experiments. Some approaches could be 1-SY projects; others would require a "team" or linkage with several disciplines.

Goal D - Research Approach D-5

Research Approach
Develop cell culture as a reproductive system for producing or maintaining new genetic combinations.

Further description of approach
Approaches will include 1) manipulation of somatic recombination; 2) manipulation of cytoplasmic recombination; 3) chromosomal reduction and manipulation; 4) interspecific gene transfer via chromosome rearrangements; 5) manipulation of intrinsic variability in order to (a) increase levels of desired variation, or (b) maintain desired stability, and (c) preserve germplasm where appropriate.

Short-term and long-term research
Both short- and long-term research included.

Disciplines or scientific expertise needed
Genetics, cytogenetics, biochemistry, physiology, pathology, and morphology.

Special facilities and equipment needed
Modern tissue culture laboratory. Other equipment as technology presents opportunities for change.

Environmental requirements
No limitation.

Immediate "research users"
Other plant scientits.

Organizational structure needed
Multidisciplinary effort, but collaborators need not be located at the same location